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APPLICATION NO.	FILING DATE	FIRST NAMED INVENTOR	ATTORNEY DOCKET NO.	CONFIRMATION NO.
08/479,997	06/07/1995	DEAN ENGELHARDT	ENZ-5(D6)(C2	8799

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EXAMINER
SPIEGLER, ALEXANDER H

ART UNIT	PAPER NUMBER
1656	47

DATE MAILED: 11/26/2001

Please find below and/or attached an Office communication concerning this application or proceeding.

Office Action Summary	Application No. 08/479,997	Applicant(s) ENGELHARDT ET AL.	
	Examiner Alexander H. Spiegler	Art Unit 1656	

-- The MAILING DATE of this communication appears on the cover sheet with the correspondence address --

Period for Reply

A SHORTENED STATUTORY PERIOD FOR REPLY IS SET TO EXPIRE 3 MONTH(S) FROM THE MAILING DATE OF THIS COMMUNICATION.

- Extensions of time may be available under the provisions of 37 CFR 1.136(a). In no event, however, may a reply be timely filed after SIX (6) MONTHS from the mailing date of this communication.
- If the period for reply specified above is less than thirty (30) days, a reply within the statutory minimum of thirty (30) days will be considered timely.
- If NO period for reply is specified above, the maximum statutory period will apply and will expire SIX (6) MONTHS from the mailing date of this communication.
- Failure to reply within the set or extended period for reply will, by statute, cause the application to become ABANDONED (35 U.S.C. § 133).
- Any reply received by the Office later than three months after the mailing date of this communication, even if timely filed, may reduce any earned patent term adjustment. See 37 CFR 1.704(b).

Status

1)☒ Responsive to communication(s) filed on *20 August 2001* .

2a)☐ This action is **FINAL**. 2b)☒ This action is non-final.

3)☐ Since this application is in condition for allowance except for formal matters, prosecution as to the merits is closed in accordance with the practice under *Ex parte Quayle*, 1935 C.D. 11, 453 O.G. 213.

Disposition of Claims

4)☒ Claim(s) *454-467* is/are pending in the application.

4a) Of the above claim(s) _____ is/are withdrawn from consideration.

5)☐ Claim(s) _____ is/are allowed.

6)☒ Claim(s) *454-467* is/are rejected.

7)☐ Claim(s) _____ is/are objected to.

8)☐ Claim(s) _____ are subject to restriction and/or election requirement.

Application Papers

9)☐ The specification is objected to by the Examiner.

10)☐ The drawing(s) filed on _____ is/are: a)☐ accepted or b)☐ objected to by the Examiner.

Applicant may not request that any objection to the drawing(s) be held in abeyance. See 37 CFR 1.85(a).

11)☐ The proposed drawing correction filed on _____ is: a)☐ approved b)☐ disapproved by the Examiner.

If approved, corrected drawings are required in reply to this Office action.

12)☐ The oath or declaration is objected to by the Examiner.

Priority under 35 U.S.C. §§ 119 and 120

13)☐ Acknowledgment is made of a claim for foreign priority under 35 U.S.C. § 119(a)-(d) or (f).

a)☐ All b)☐ Some * c)☐ None of:

1.☐ Certified copies of the priority documents have been received.

2.☐ Certified copies of the priority documents have been received in Application No. _____ .

3.☐ Copies of the certified copies of the priority documents have been received in this National Stage application from the International Bureau (PCT Rule 17.2(a)).

* See the attached detailed Office action for a list of the certified copies not received.

14)☐ Acknowledgment is made of a claim for domestic priority under 35 U.S.C. § 119(e) (to a provisional application).

a) ☐ The translation of the foreign language provisional application has been received.

15)☒ Acknowledgment is made of a claim for domestic priority under 35 U.S.C. §§ 120 and/or 121.

Attachment(s)

1) ☒ Notice of References Cited (PTO-892)
2) ☐ Notice of Draftsperson's Patent Drawing Review (PTO-948)
3) ☐ Information Disclosure Statement(s) (PTO-1449) Paper No(s) _____ .
4) ☐ Interview Summary (PTO-413) Paper No(s). _____ .
5) ☐ Notice of Informal Patent Application (PTO-152)
6) ☐ Other: .

DETAILED ACTION

1. In view of the appeal brief, filed 8/20/01, and newly found rejections summarized herein, PROSECUTION IS HEREBY REOPENED. New grounds of rejection are set forth below. To avoid abandonment of the application, appellant must exercise one of the following two options: 1) file a reply under 37 CFR 1.111; or 2) request reinstatement of the appeal. If reinstatement of the appeal is requested, such request must be accompanied by a supplemental appeal brief, but no new amendments, affidavits (37 CFR 1.30, 1.131, or 1.132) or other evidence are permitted. See 37 CFR 1.93 (b) (2).

It is noted that the amended claims submitted in the Appeal Brief have been entered and have been examined on the merits, therefore, currently amended claims 454-567 are pending. Any objections and rejections not reiterated below are hereby withdrawn.

Claim Rejections - 35 USC § 112

2. The following is a quotation of the first paragraph of 35 U.S.C. 112:

> The specification shall contain a written description of the invention, and of the manner and process of making and using it, in such full, clear, concise, and exact terms as to enable any person skilled in the art to which it pertains, or with which it is most nearly connected, to make and use the same and shall set forth the best mode contemplated by the inventor of carrying out his invention.

3. Claims 461, 489, 518, and 546 rejected under 35 U.S.C. 112, first paragraph, as containing subject matter which was not described in the specification in such a way as to reasonably convey to one skilled in the relevant art that the inventor(s), at the time the application was filed, had possession of the claimed invention. These claims include the recitation of "wherein chemical linkage comprises or includes an olefinic bond at the delta-position relative to the point of attachment" which does not appear in the specification. This recitation is considered new matter.

4. The following is a quotation of the second paragraph of 35 U.S.C. 112:

> The specification shall conclude with one or more claims particularly pointing out and distinctly claiming the subject matter which the applicant regards as his invention.

5. Claims 455, 483, 512, and 540 rejected under 35 U.S.C. 112, second paragraph, as being indefinite for failing to particularly point out and distinctly claim the subject matter which applicant regards as the invention.

A) Claims 455, 483, 512, and 540 are indefinite over the recitation of "self-signaling or self-indicating or self-detecting" because it is not clear what is meant by this recitation. (i.e. it is not clear as to how a Sig can be considered be self-signaling or self-indicating or self-detecting). For example, a fluorescent compound needs a specific wavelength of light to excite the compound to fluoresce and optical detection system to detect emitted fluorescence, therefore it is not clear as to how a Sig (for example a fluorescent compound) could be self-signaling or self-indicating or self-detecting. In other words, it is not clear as to how a Sig can be considered self-signaling or self-indicating or self-detecting without the use of an additional element to aid in the signaling, indicating or detecting of the Sig.

Claim Rejections - 35 USC § 102

6. The following is a quotation of the appropriate paragraphs of 35 U.S.C. 102 that form the basis for the rejections under this section made in this Office action:

> A person shall be entitled to a patent unless –
>
> (e) the invention was described in a patent granted on an application for patent by another filed in the United States before the invention thereof by the applicant for patent, or on an international application by another who has fulfilled the requirements of paragraphs (1), (2), and (4) of section 371(c) of this title before the invention thereof by the applicant for patent.

7. Claims 454-461, 463-474, 476-489, 491-502, 504-518, 520-531, 533-546, 548-559, and 561-567 are rejected under 35 U.S.C. 102(e) as being anticipated by Ward et al. (USPN 4,711,955).

Ward teaches modified nucleotides and methods of using and preparing the same. Specifically, Ward teaches the production and use of nucleic acid probes comprising a general structure (see abstract),

"wherein B represents a purine, 7-deazapurine, or pyrimidine moiety covalently bonded to the C1' -position of the sugar moiety, provided that when B is purine or 7-deazapurine, it is attached at the N9 -position of the purine or 7-deazapurine and when B is pyrimidine, it is attached at the N1 -position; wherein A represents a moiety (i.e. Sig) consisting of at least three carbon atoms which is capable of forming a detectable complex with a polypeptide when the compound is incorporated into a double-stranded ribonucleic acid, deoxyribonucleic acid duplex, or DNA-RNA hybrid; wherein the dotted line represents a chemical linkage joining B and A, provided that if B is purine, the linkage is attached to the 8-position of the purine, if B is 7-deazapurine, the linkage is attached to the 7-position of the deazapurine, and if B is pyrimidine, the linkage is attached to the 5-position of the pyrimidine and wherein each of x, y and z represents (H-, HO-, etc. see abstract) either directly, or when incorporated into oligo- and polynucleotides, provide probes which are widely useful." (see abstract).

It is noted that the claims of the instant invention are broadly drawn to oligos- or polydeoxynucleotides or polyribonucleotides, wherein the Sig is covalently attached to the PM (or x, y, or Z) directly ***or through a chemical linkage***. Ward teaches the covalent attachment of the Sig to the PM through a chemical linkage (i.e. through the linkage of the sugar (SM) and the base (BASE) moieties – see abstract), therefore, Ward teaches instant claims 454, 482, 510, 511, 539, and 567, and claims 457, 485, 514, and 542.

With respect to claims 455-56, 458, 463-474, 476-481, 483-484, 486, 491-502, 504-509, 512-513, 515, 520-531, 533-538, 540-541, 543, 548-559, 561-566 the reference teaches that the probe (for general formula, see abstract) comprising the Sig can be a self-signalling moiety such

as biotin, fluorescent dyes, electron-dense reagents (such as ferritin, colloidal gold, ferric oxide), or enzymes (such as peroxidase, alkaline phosphatase) (col. 18, 9-68).

With respect to claims 459-461, 487-489, 516-518, and 544-546, Ward teaches:

"the chemical linkages may include *any of the well known bonds including carbon-carbon single bonds, carbon-carbon double bonds, carbon-nitrogen single bonds, or carbon-oxygen single bonds*. However, it is generally preferred that *the chemical linkage include an olefinic bond at the .alpha.-position relative to B*. The presence of such an .alpha.-olefinic bond serves to hold the moiety A away from the base when the base is paired with another in the well known double-helix configuration. This permits interaction with polypeptide to occur more readily, thereby facilitating complex formation. Moreover, single bonds with greater rotational freedom may not always hold the moiety sufficiently apart from the helix to permit recognition by and complex formation with polypeptide. *It is even more preferred that the chemical linkage group be derived from a primary amine, and have the structure --CH.sub.2 --NH--*, since such linkages are easily formed utilizing any of the well known amine modification reactions. *Examples of preferred linkages derived from allylamine and allyl-(3-amino-2-hydroxyl-1-propyl) ether groups have the formulae --CH.dbd.CH--CH.sub.2 --NH-- and ##STR12## respectively.*
Although these linkages are preferred, others can be used, including particularly olefin linkage arms with other modifiable functionalities such as thiol, carboxylic acid, and epoxide functionalities.

This rejection could be overcome by amending the claims by deleting the recitation *"or through a chemical linkage".*

8. Claims 454-459, 463-464, 472-475, 478-487, 491-492, 500-503, and 506-510 are rejected under 35 U.S.C. 102(b) as anticipated by or, in the alternative, under 35 U.S.C. 103(a) as obvious over Halloran et al. (J. of Immun. (1966), 96(3): 373-378).

Halloran teaches a Sig label (i.e. a protein) attached to nucleic acids (pg. 373, 374 - Fig.1 and col. 2). Specifically, Halloran teaches the preparation of nucleotide protein conjugates through the covalent linkage of a protein to a nucleotide (on the phosphate moiety) with a carbodiimide coupling agent (Fig. 1). Halloran also teaches that this conjugation can be applied to mononucleotides, oligonucleotides, and DNA (pg. 373, col. 1). Therefore,

Halloran teaches an oligonucleotide, comprising at least one modified nucleotide having the formula Sig – PM – SM – BASE, wherein the Sig is covalently attached to the PM directly, said Sig being a moiety capable of non-radioactive detection (i.e. instant claims 454, 481, 482, 509, and 510).

With respect to claims 455 and 483, Halloran teaches that the Sig is a protein, which is or renders the nucleotide self-signaling or self-indicating or self-detecting. Halloran teaches the detection of the protein and DNA through Amidoschwartz and Feulgan staining, respectively (pg. 374, col. 2).

With respect to claims 456, 458, 464, 472-475, 478, 479, 484, 486, 492, 500-503, and 506-508 Halloran teaches the addition of proteins (such as HSA and poly-lysine) (pg. 375, Table 1), which comprise at least three carbon atoms.

With respect to claims 457, 459, 463, 480, 485, 487, 491, and 508 Halloran teaches the covalent attachment of –P–O– , said chemical linkage of -CH2NH- , and where the Sig is covalently attached to the PM through a phosphorous atom or phosphate oxygen (pg. 374, Fig. 1).

Therefore, even though Halloran does not teach the hybridization of an oligodeoxynucleotide to a nucleic acid of interest, or a portion thereof, but it is an inherent property of an oligonucleotide to hybridize to a nucleic acid of interest (or portion thereof), which is complementary to the oligonucleotide.

It is noted that in In re Best (195 USPQ 430) and In re Fitzgerald (205 USPQ 594) discuss the support of rejections wherein the prior art discloses subject matter which there is reason to believe inherently includes functions that are newly cited or is identical to a product

instantly claimed. In such a situation the burden is shifted to the applicants to "prove that subject matter shown to be in the prior art does not possess characteristic relied on" (205 USPQ 594, second column, first full paragraph).

In the alternative, it would have been obvious to one of ordinary skill in the art at the time the invention was made to have used the oligonucleotide preparation method of Halloran of conjugating a protein to an oligonucleotide (through the PM), in order to have produced a compound that was complementary to a nucleic acid of interest for detection and identification purposes. If the hybridization property of oligonucleotides is not inherent, the disclosure of oligonucleotides, per se, suggests hybridizability, which is a well known characteristic. Therefore, the burden is shifted to the applicant to prove this otherwise.

9. Claims 511-516, 520-521, 529-532, 535, 536-544, 548, 549, 557-560, and 563-567 are rejected under 35 U.S.C. 102(b) as anticipated by or, in the alternative, under 35 U.S.C. 103(a) as obvious over Halloran et al. (J. of Immun. (1966), 96(3): 379-385).

Halloran teaches the conjugation of proteins to mono, oligo, and polynucleotides (pg. 379, i.e. reference to preceding article - Halloran et al. (J. of Immun. (1966), 96(3): 373-378), see teachings above. The teachings of Halloran (pgs. 373-378) are cited herein only to demonstrate content of Halloran (379-385)).

Halloran (pg. 381, colum 2) teaches:

"The results of the immunologic studies indicate that nucleotides, oligonucleotides and DNA-protein conjugates induce can induce the formation of antibodies with nucleotide specificity. The antibodies react both with denatured DNA and with nucleotide protein conjugates. *While immunologic response to analagous RNA protein preparations has not been studied, it may be presumed that antibodies to the different types of RNA could be obtained by the same procedure*".

Therefore, Halloran teaches an oligoribonucleotide, comprising at least one modified nucleotide having the formula Sig – PM – SM – BASE, wherein the Sig is covalently attached to the PM directly, said Sig being a moiety capable of non-radioactive detection (i.e. instant claims 511, 538, 539, 566, and 567).

With respect to claims 512 and 540, Halloran teaches that the Sig is a protein, which is or renders the nucleotide self-signaling or self-indicating or self-detecting. Halloran teaches the detection of the protein and DNA through Amidoschwartz and Feulgan staining, respectively (pg. 374, col. 2).

With respect to claims 513, 515, 521, 529-532, 535-536, 541, 543, 549, 557-560, 563, and 564 Halloran teaches the addition of proteins (such as HSA and poly-lysine) (pg. 375, Table 1), which comprise at least three carbon atoms.

With respect to claims 514, 516, 520, 537, 542, 544, 548, and 565 Halloran teaches the covalent attachment of –P–O– , said chemical linkage of -CH2NH- , and where the Sig is covalently attached to the PM through a phosphorous atom or phosphate oxygen (pg. 374, Fig. 1).

Therefore, even though Halloran does not teach the hybridization of an oligoribonucleotide to a nucleic acid of interest, or a portion thereof, but it is an inherent property of an oligoribonucleotide to hybridize to a nucleic acid of interest (or portion thereof), which is complementary to the oligonucleotide.

It is noted that in In re Best (195 USPQ 430) and In re Fitzgerald (205 USPQ 594) discuss the support of rejections wherein the prior art discloses subject matter which there is reason to believe inherently includes functions that are newly cited or is identical to a product

instantly claimed. In such a situation the burden is shifted to the applicants to "prove that subject matter shown to be in the prior art does not possess characteristic relied on" (205 USPQ 594, second column, first full paragraph).

In the alternative, in view of the teachings of Halloran (pgs. 379-385) it would have been obvious to one of ordinary skill in the art at the time the invention was made to have modified the method of Halloran (pgs. 373-378) so as to have conjugated a protein to an RNA molecule, in order to have achieved an equally effective compound for use in hybridization or antibody production. If the hybridization property of oligoribonucleotides is not inherent, the disclosure of oligoribonucleotides, per se, suggests hybridizability, which is a well known characteristic. Therefore, the burden is shifted to the applicant to prove this otherwise.

Claim Rejections - 35 USC § 103

10. The following is a quotation of 35 U.S.C. 103(a) which forms the basis for all obviousness rejections set forth in this Office action:

> (a) A patent may not be obtained though the invention is not identically disclosed or described as set forth in section 102 of this title, if the differences between the subject matter sought to be patented and the prior art are such that the subject matter as a whole would have been obvious at the time the invention was made to a person having ordinary skill in the art to which said subject matter pertains. Patentability shall not be negatived by the manner in which the invention was made.

11. Claims 462, 464, 469-471, 476, 477, 490, 492, 497-499, 504, and 505 are rejected under 35 U.S.C. 103(a) as being unpatentable over Halloran et al. (J. of Immun. (1966), 96(3): 373-378), as applied to claims 454-459, 463-464, 472-475, 478-487, 491-492, 500-503, and 506-510 above, and further in view of Falkow et. al (USPN 4,358,535).

The teachings of Halloran are presented above. Specifically, Halloran teaches an oligonucleotide, comprising at least one modified nucleotide having the formula Sig – PM – SM

– BASE, wherein the Sig is covalently attached to the PM directly, said Sig being a moiety capable of non-radioactive detection (i.e. protein). Halloran also describes specific linkages and attachments of the Sig and the PM. Halloran does not teach specific Sig's, such as fluorescent compounds, ligands, etc. of nucleic acid probes.

Falkow teaches methods and compositions for infectious disease diagnosis and epidemiology involving labeled nucleotide probes complementary to nucleic acid coding for a characteristic pathogen product (see abstract). Specifically, Falkow teaches:

"The probe may be RNA or DNA. The probe will normally have at least about 25 bases, more usually at least about 30 bases, and may have up to about 10,000 bases or more, usually having not more than about 5,000 bases. The probe sequence will be at least substantially complementary to a gene coding for a product characteristic of the pathogen, usually a cytoplasmic product or released product, particularly an excreted product. The probe need not have perfect complementarity to the sequence to which it hybridizes; there may by 30% or more of mismatched pairs." (col. 2, ln. 42-54).

With respect to claims 462, 469, 490, and 497 Falkow teaches that enzymes of interest as labels (i.e. Sigs) include hydrolases, particularly esterases and glycosidases (i.e. which would be attached to the PM via a glycosidic linkage), or oxidoreductases, particularly peroxidases (col. 4, ln. 12-14).

With respect to claims 471 and 499, the reference teaches that fluorescent compounds include fluorescein and its derivatives, rhodamine and its derivatives, dansyl, umbelliferone, etc. (col. 4, ln. 14-16).

With respect to claims 470 and 498, Falkow teaches that the probe can be labeled with heavy metals (i.e. which are catalytic) (col. 3, ln. 25-28).

With respect to claims 476-477 and 504-505, Falkow teaches:

"In some situations it may be feasible to employ an antibody (i.e. a polypeptide) which will bind specifically to the probe hybridized to the single stranded DNA of the pathogen. In this

instance, the antibody would be labeled to allow for detection. The same types of labels which are used for the probe may also be bound to the antibody in accordance with known techniques." (col. 3, ln. 28-34).

"Other labels include ligands, which can serve as a specific binding member to a labeled antibody, fluorescers, chemiluminescers, enzymes, antibodies which can serve as a specific binding pair member for a labeled ligand, and the like. A wide variety of labels have been employed in immunoassays which can readily be employed in the present assay." (col. 3, ln. 38-45).

"Ligands and antiligands may be varied widely. Where a ligand has a natural receptor, namely ligands such as biotin, thyroxine, and cortisol, these ligands can be used in conjunction with labeled naturally occurring receptors. Alternatively, any compound can be used, either haptenic or antigenic, in combination with an antibody." (col. 4, ln. 5-11).

In view of the teachings of Falkow, it would have been obvious to one of ordinary skill in the art at the time the invention was made to have modified the oligonucleotides of Halloran so as to have used alternative Sig labels, instead of using a protein, in order to have provided an equally effective compound for nucleic acid detection.

12. Claims 519, 521, 526-528, 533, 534, 547, 549, 554-556, 561, and 562 are rejected under 35 U.S.C. 103(a) as being unpatentable over Halloran et al. (J. of Immun. (1966), 96(3): 379-385), as applied to claims 511-516, 520-521, 529-532, 535, 536-544, 548, 549, 557-560, and 563-567 above, and further in view of Falkow et. al (USPN 4,358,535).

The teachings of Halloran are presented above. Specifically, Halloran teaches an oligoribonucleotide, comprising at least one modified nucleotide having the formula Sig – PM – SM – BASE, wherein the Sig is covalently attached to the PM directly, said Sig being a moiety capable of non-radioactive detection (i.e. protein). Halloran also describes specific linkages and attachments of the Sig and the PM. Halloran does not teach specific Sig's, such as fluorescent compounds, ligands, etc. of nucleic acid probes.

Falkow teaches methods and compositions for infectious disease diagnosis and epidemiology involving labeled nucleotide probes complementary to nucleic acid coding for a characteristic pathogen product (see abstract). Specifically, Falkow teaches:

"The probe may be RNA or DNA. The probe will normally have at least about 25 bases, more usually at least about 30 bases, and may have up to about 10,000 bases or more, usually having not more than about 5,000 bases. The probe sequence will be at least substantially complementary to a gene coding for a product characteristic of the pathogen, usually a cytoplasmic product or released product, particularly an excreted product. The probe need not have perfect complementarity to the sequence to which it hybridizes; there may by 30% or more of mismatched pairs." (col. 2, ln. 42-54).

With respect to claims 519, 526, 547, and 554 Falkow teaches that enzymes of interest as labels (i.e. Sigs) include hydrolases, particularly esterases and glycosidases (i.e. which would be attached to the PM via a glycosidic linkage), or oxidoreductases, particularly peroxidases (col. 4, ln. 12-14).

With respect to claims 528 and 556, the reference teaches that fluorescent compounds include fluorescein and its derivatives, rhodamine and its derivatives, dansyl, umbelliferone, etc. (col. 4, ln. 14-16).

With respect to claims 527 and 555, Falkow teaches that the probe can be labeled with heavy metals (i.e. which are catalytic) (col. 3, ln. 25-28).

With respect to claims 533-534 and 561-562, Falkow teaches:

"In some situations it may be feasible to employ an antibody (i.e. a polypeptide) which will bind specifically to the probe hybridized to the single stranded DNA of the pathogen. In this instance, the antibody would be labeled to allow for detection. The same types of labels which are used for the probe may also be bound to the antibody in accordance with known techniques." (col. 3, ln. 28-34).

"Other labels include ligands, which can serve as a specific binding member to a labeled antibody, fluorescers, chemiluminescers, enzymes, antibodies which can serve as a specific binding pair member for a labeled ligand, and the like. A wide variety of labels have been

employed in immunoassays which can readily be employed in the present assay." (col. 3, ln. 38-45).

"Ligands and antiligands may be varied widely. Where a ligand has a natural receptor, namely ligands such as biotin, thyroxine, and cortisol, these ligands can be used in conjunction with labeled naturally occurring receptors. Alternatively, any compound can be used, either haptenic or antigenic, in combination with an antibody." (col. 4, ln. 5-11).

In view of the teachings of Falkow, it would have been obvious to one of ordinary skill in the art at the time the invention was made to have modified the oligonucleotides of Halloran so as to have used alternative Sig labels, instead of using a protein, in order to have provided an equally effective compound for nucleic acid detection.

13. Claims 460-461, 465-468, 488-489, and 493-496 are rejected under 35 U.S.C. 103(a) as being unpatentable over Halloran et al. (J. of Immun. (1966), 96(3): 373-378), as applied to claims 454-459, 463-464, 472-475, 478-487, 491-492, 500-503, and 506-510 above, and further in view of Ward et. al (USPN 4,711,955).

The teachings of Halloran are presented above. Specifically, Halloran teaches an oligonucleotide, comprising at least one modified nucleotide having the formula Sig – PM – SM – BASE, wherein the Sig is covalently attached to the PM directly, said Sig being a moiety capable of non-radioactive detection (i.e. protein). Halloran also describes specific linkages and attachments of the Sig and the PM. Halloran does not teach the chemical linkages comprising an allylamine group or an ether group and Halloran does not teach a Sig comprising ferritin.

Ward teaches that the probe (for general formula, see abstract) comprising the Sig can be a self-signalling moiety such as biotin, fluorescent dyes, electron-dense reagents (such as ferritin, colloidal gold, ferric oxide), or enzymes (such as peroxidase, alkaline phosphatase) (col. 18, 9-68).

With respect to specific linkages, Ward teaches:

"the chemical linkages may include *any of the well known bonds including carbon-carbon single bonds, carbon-carbon double bonds, carbon-nitrogen single bonds, or carbon-oxygen single bonds*. However, it is generally preferred that *the chemical linkage include an olefinic bond at the .alpha.-position relative to B*. The presence of such an .alpha.-olefinic bond serves to hold the moiety A away from the base when the base is paired with another in the well known double-helix configuration. This permits interaction with polypeptide to occur more readily, thereby facilitating complex formation. Moreover, single bonds with greater rotational freedom may not always hold the moiety sufficiently apart from the helix to permit recognition by and complex formation with polypeptide. *It is even more preferred that the chemical linkage group be derived from a primary amine, and have the structure --CH.sub.2 --NH--*, since such linkages are easily formed utilizing any of the well known amine modification reactions. *Examples of preferred linkages derived from allylamine and allyl-(3-amino-2-hydroxyl-1-propyl) ether groups have the formulae --CH.dbd.CH--CH.sub.2 --NH-- and ##STR12## respectively.*

Although these linkages are preferred, others can be used, including particularly olefin linkage arms with other modifiable functionalities such as thiol, carboxylic acid, and epoxide functionalities.

It is also noted that the instant specification teaches that the "Sig component of the nucleotides in accordance with this invention and the nucleotides and polynucleotides incorporating the nucleotides of this invention containing the Sig component are equivalent to and useful for the same purposes as the nucleotides described in U.S. patent application Serial No. 255,223, now U.S. Patent NO. 4,711,955". (pg. 97).

In view of the teachings of Ward, it would have been obvious to one of ordinary skill in the art at the time the invention was made to have modified the method of linkage of Halloran to include allylamine, ether, or any other well known chemical linkage, instead of a carbodiimide linkage as taught by Halloran, so as to have achieved an equally effective linkage between the Sig and the phosphate moiety. Furthermore, one would have been motivated to use the Sig labels of Ward (i.e. ferritin), instead of using a protein, in order to have provided an equally effective compound for nucleic acid detection.

14. Claims 517-518, 522-525, 545-546, and 550-553 are rejected under 35 U.S.C. 103(a) as being unpatentable over Halloran et al. (J. of Immun. (1966), 96(3): 379-385), as applied to claims 511-516, 520-521, 529-532, 535, 536-544, 548, 549, 557-560, and 563-567 above, and further in view of Ward et. al (USPN 4,711,955)

The teachings of Halloran are presented above. Specifically, Halloran teaches an oligoribonucleotide, comprising at least one modified nucleotide having the formula Sig – PM – SM – BASE, wherein the Sig is covalently attached to the PM directly, said Sig being a moiety capable of non-radioactive detection (i.e. protein). Halloran also describes specific linkages and attachments of the Sig and the PM. Halloran does not teach the chemical linkages comprising an allylamine group or an ether group and Halloran does not teach a Sig comprising ferritin.

Ward teaches that the probe (for general formula, see abstract) comprising the Sig can be a self-signalling moiety such as biotin, fluorescent dyes, electron-dense reagents (such as ferritin, colloidal gold, and ferric oxide), or enzymes (such as peroxidase, alkaline phosphatase) (col. 18, 9-68).

With respect to specific linkages, Ward teaches:

"the chemical linkages may include *any of the well known bonds including carbon-carbon single bonds, carbon-carbon double bonds, carbon-nitrogen single bonds, or carbon-oxygen single bonds*. However, it is generally preferred that *the chemical linkage include an olefinic bond at the .alpha.-position relative to B*. The presence of such an .alpha.-olefinic bond serves to hold the moiety A away from the base when the base is paired with another in the well known double-helix configuration. This permits interaction with polypeptide to occur more readily, thereby facilitating complex formation. Moreover, single bonds with greater rotational freedom may not always hold the moiety sufficiently apart from the helix to permit recognition by and complex formation with polypeptide. *It is even more preferred that the chemical linkage group be derived from a primary amine, and have the structure --CH.sub.2 --NH--*, since such linkages are easily formed utilizing any of the well known amine modification reactions. *Examples of preferred linkages derived from allylamine and allyl-(3-amino-2-hydroxyl-1-propyl) ether groups have the formulae --CH.dbd.CH--CH.sub.2 --NH-- and ##STR12## respectively.*

Although these linkages are preferred, others can be used, including particularly olefin linkage arms with other modifiable functionalities such as thiol, carboxylic acid, and epoxide functionalities.

It is also noted that the instant specification teaches that the "Sig component of the nucleotides in accordance with this invention and the nucleotides and polynucleotides incorporating the nucleotides of this invention containing the Sig component are equivalent to and useful for the same purposes as the nucleotides described in U.S. patent application Serial No. 255,223, now U.S. Patent NO. 4,711,955". (pg. 97).

In view of the teachings of Ward, it would have been obvious to one of ordinary skill in the art at the time the invention was made to have modified the method of linkage of Halloran to include allylamine, ether, or any other well known chemical linkage, instead of a carbodiimide linkage as taught by Halloran, so as to have achieved an equally effective linkage between the Sig and the phosphate moiety. Furthermore, one would have been motivated to use the Sig labels of Ward (i.e. ferritin), instead of using a protein, in order to have provided an equally effective compound for nucleic acid detection.

15. Claims 475 and 503 are rejected under 35 U.S.C. 103(a) as being unpatentable over Ward et al. (USPN 4,711,955) as applied to claims 454-461, 463-474, 476-489, 491-502, 504-518, 520-531, 533-546, 548-559, and 561-567 above, and further in view of Halloran et al. (J. of Immun. (1966), 96(3): 373-378).

The teachings of Ward are presented above. Specifically, Ward teaches the probe comprising Sig – PM – SM – BASE, wherein the covalent attachment of the Sig to the PM through a chemical linkage (i.e. through the linkage of the sugar (SM) and the base (BASE) moieties – see

abstract). Ward also teaches that a variety of Sig labels can be used in detection (col. 18, ln. 21-28), but does not teach the Sig label comprising polylysine.

The teachings of Halloran are presented above. Specifically, Halloran teaches the addition of proteins (such as HSA and poly-lysine) to the PM, which can be used in detection with the Amidoschwartz staining procedure, for example (referenced to the pg. 375, Table 1 – referenced).

In view of the teachings of Halloran, it would have been obvious to one of ordinary skill in the art at the time the invention was made to have modified the compound of Ward to include to a Sig comprising polylysine, instead of a Sig (such as rhodamine), so as to have achieved an equally effective compound for nucleic acid detection.

16. Claims 532 and 560 are rejected under 35 U.S.C. 103(a) as being unpatentable over Ward et al. (USPN 4,711,955) as applied to claims 454-461, 463-474, 476-489, 491-502, 504-518, 520-531, 533-546, 548-559, and 561-567 above, and further in view of Halloran et al. (J. of Immun. (1966), 96(3): 379-385).

The teachings of Ward are presented above. Specifically, Ward teaches the probe comprising Sig – PM – SM – BASE, wherein the covalent attachment of the Sig to the PM through a chemical linkage (i.e. through the linkage of the sugar (SM) and the base (BASE) moieties – see abstract). Ward also teaches that a variety of Sig labels can be used in detection (col. 18, ln. 21-28), but does not teach the Sig label comprising polylysine.

The teachings of Halloran are presented above. Specifically, Halloran teaches the addition of proteins (such as HSA and poly-lysine) to the PM of a oligoribonucleotide, which can be used in detection with the Amidoschwartz staining procedure, for example (pg. 379, i.e.

reference to preceding article - Halloran et al. (J. of Immun. (1966), 96(3): 373-378)).

In view of the teachings of Halloran, it would have been obvious to one of ordinary skill in the art at the time the invention was made to have modified the compound of Ward to include to a Sig comprising polylysine, instead of a Sig (such as rhodamine), so as to have achieved an equally effective compound for nucleic acid detection.

17. Claims 462, 490, 519, and 547 are rejected under 35 U.S.C. 103(a) as being unpatentable over Ward et al. (USPN 4,711,955) as applied to claims 454-461, 463-474, 476-489, 491-502, 504-518, 520-531, 533-546, 548-559, and 561-567 above, and further in view of Falkow et al. 4,358,535.

The teachings of Ward are presented above. Specifically, Ward teaches the probe comprising Sig – PM – SM – BASE, wherein the covalent attachment of the Sig to the PM through a chemical linkage (i.e. through the linkage of the sugar (SM) and the base (BASE) moieties – see abstract). Ward also teaches that a variety of Sig labels (such as peroxidase and alkaline phosphatase) can be used in detection (col. 18, ln. 24-28), but does not teach the Sig label comprising a glycosidic linkage (i.e. using a Sig comprising a glycosidase).

The teachings of Falkow are presented above. Specifically, Falkow teaches enzymes of interest as labels (i.e. Sigs) include hydrolases, particularly esterases and glycosidases (i.e. which would be attached to the PM via a glycosidic linkage), or oxidoreductases, particularly peroxidases (col. 4, ln. 12-14).

In view of the teachings of Falkow, it would have been obvious to one of ordinary skill in the art at the time the invention was made to have modified the compound of Ward to include to a Sig comprising a glycosidic linkage (i.e. a Sig comprising a glycosidase), instead of a Sig (such

as peroxidase, utilizing a different linkage), so as to have achieved an equally effective compound for nucleic acid detection.

18. **No claims are allowable.**

Any inquiry concerning this communication or earlier communications from the examiner should be directed to Alexander H. Spiegler whose telephone number is (703) 305-0806. The examiner can normally be reached on Monday through Friday, 7:00 AM to 3:30 PM.

If attempts to reach the examiner by telephone are unsuccessful, the examiner's supervisor, W. Gary Jones can be reached on (703) 308-1152. The fax phone numbers for the organization where this application or proceeding is assigned are (703) 308-4242 and (703) 305-3014.

Any inquiry of a general nature or relating to the status of this application or proceeding should be directed to the receptionist whose telephone number is (703) 308-0196.

Alexander H. Spiegler
November 19, 2001

W. Gary Jones
Supervisory Patent Examiner
Technology Center 1600